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File: USPT

Feb 2, 1999

DOCUMENT-IDENTIFIER: US 5866328 A

TITLE: Fast DNA sequence determination method by measuring energy of base pairing or unpairing of nucleic acid sequences and use thereof in sequencing and diagnostics

BSPR:

The simplest is to functionalize, using synthetic oligonucleotides, one of the ends of a double-stranded DNA with two different functions (biotin and amine, for example), which permit anchoring to two different pre-treated surfaces. The two strands at the other end may be joined using a partially paired synthetic nucleotide in the form of a loop. In this way, a paired, single-stranded DNA is produced from a double-stranded DNA (see FIG. 2(a)). The advantage of this method lies in its capacity to functionalize a heterogeneous population of large DNA fragments (as are obtained by fractionation of a gene or chromosome), which can then be analyzed simultaneously. In this case, the DNA sample is fractionated using two (or more) restriction enzymes, which enables a subpopulation to be obtained with two different restriction sites at its ends which are similar over all the fragments. This enables the two ends to be treated differently (for example by joining one of end to an oligonucleotide in the form of a loop possessing the appropriate restriction site at its end). The drawback of this method lies in the steric interference between the two adjacent functional groups, which can make coupling to the surfaces difficult.

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